Application No. 10/536,533

Paper Dated: September 4, 2008

In Reply to USPTO Correspondence of June 4, 2008

Attorney Docket No. 4544-051675

AMENDMENTS TO THE SPECIFICATION

Please amend the specification accordingly:

Please amend the paragraph beginning on page 6, line 22 as follows:

-- To the suspension of latex particles prepared in step (b), 0.6-1.0 mg preferably 0.8 mg per ml of the suspension, immunolobulins immunoglobulins prepared in step (a) are added. The whole mixture is then rotated end-over-end for 18-20 hours at a temperature of 20-25°C. The coating reaction is stopped by addition of 1M glycine (pH 11.0) taken in quantity of 0.06 ml per ml of solution of immunoglobulin coated latex particles. Rotation is continued for about 30 minutes at a temperature of 20-25°C. The coated latex particles are pelleted out by centrifugation at 10,000 rpm for 10-12 minutes at a temperature of about 4°C. The pellet is washed thrice with washing buffer (50 mM glycine, pH 8.5; 0.03% TRITON triton X-100 m and 0.05% sodium azide) at 10,000 rpm for 10-12 minutes at a temperature of about 4°C. Finally the washed coated latex particles are resuspended in storage buffer (50 mM glycine, pH 8.5; 1.0% bovine serum albumin; 0.03% TRITON triton—X-100 m; 0.1% sodium azide and 0.01% thiomersol to a final concentration of 1% and sonicated by a tip sonicator for around 60 seconds at about 5 watts and stored at 4°C. --

Please amend the paragraph beginning on page 8, line 8 as follows:

-- Flagellin gene sequence specific to salmonella typhi was amplified by polymerase chain reaction (PCR) using gene specific primers. Amplified PCR product was cloned in Glutathione-S-transferase (GST) vector and later expressed. The expressed protein was purified by GST affinity column chromatography. The protein content of the purified product was determined by the Bradford method. Hyper immune serum against this protein was raised in rabbit. Immunoglobulins fraction of hyper immune sera was separated by ammonium sulphate precipitation. The precipitated immunoglobulins were suspended in 1.0 ml PB (50 mM, pH 7.2), dialysed and protein content determined. 1.0 ml of 1% carboxylated latex particles and 1.0 ml of 40 mM MES buffer (pH 5.5-6.5) were taken in 2.0 ml microcentrifuge tube. Then they were

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mixed on vortex mixer for 60 seconds and centrifuged at 10,000 RPM for 10-12 minutes at a temperature of 4°C. The latex particles were further washed twice in 2.0 ml of 20 mM MES buffer (pH 5.5) by mixing on vortex mixer for 60 seconds and centrifugation at 10,000 RPM for 10-12 minutes at a temperature of 4°C. Following the final wash, the latex particles were suspended in 1.0 ml MES buffer (20 mM, pH 5.5) and sonicated by a tip sonicator at 5 watts for 60-120 seconds. Later 1.0 ml of freshly prepared solution of 0.1 M 1-ethyl-3-(3dimethylaminopropyl)carbodimide hydrochloride (EDC) in MES buffer (20 mM, pH 5.5) was added drop wise while the solution was slowly vortexed. Then the tube was rotated slowly endover-end for 3 hours at a temperature of 20-25°C followed by washing three times with MES buffer (20 mM, pH 5.5) at 10,000 RPM for 10-12 minutes at a temperature of 4°C. The latex particles were resuspended in 0.7 ml MES buffer (20 mM, pH 5.5) and sonicated for 60-120 seconds by a tip sonicator at 5 watts. 0.8 mg of immunoglobulins were added to latex particles and volume was made up to 1.0 ml with MES buffer (20 mM, pH 5.5). This was then rotated end-over-end for 18-20 hours at a temperature of 20-25°C. The coating reaction was then stopped by addition of 0.06 ml of 1M glycine (pH 11.0). The rotation was continued for 30 minutes at a temperature of 20-25°C. The coated latex particles were pelleted out by centrifugation at 10,000 RPM for 10-12 minutes at a temperature of 4°C. The pellet was washed thrice with 2.0 ml of washing buffer (50 mM glycine, pH 8.5, 0.03% TRITON triton-X-100™ and 0.05% sodium azide) at 10,000 RPM for 10-12 minutes at a temperature of 4°C. The washed coated latex particles were resuspended in storage buffer (50 mM glycine, pH 8.6. 1.0% bovine serum albumin, 0.03% TRITONtriton X-100TM, 0.1% sodium azide and 0.01% thiomersol) to a final concentration of 1% and sonicated with tip sonicator for 60 seconds at 5 watts and stored at 4°C.